

**APPENDIX A: Mark-up of Amended Claims**

1. (amended) A method for producing a vaccine composition containing an immunogenic determinant as the active ingredient, [characterised in that] the method comprising [comprises] the steps of:
  - a. treating procaryotic cells under conditions such that an increase of the concentration of trehalose within the procaryotic cells is induced;
  - b. using the induced cells containing trehalose as the immunogenic determinant in the production of a vaccine composition.
2. (amended) [A] The method as claimed in claim 1, [characterised in that] wherein the treatment of the procaryotic cells is carried out to achieve a concentration of trehalose within the cells of at least 10 mM.
3. (amended) [A] The method as claimed in claim 1, wherein [either of claims 1 or 2, characterised in that] the increase in concentration of trehalose is achieved by synthesis of trehalose within the cell.
4. (amended) [A] The method as claimed in claim 1, wherein [any one of the preceding claims, characterised in that] the condition causing the increase of trehalose concentration within the cells is heat, osmotic shock, suppression of degradation of trehalose, or genetically engineered constitutive synthesis of trehalose within the cells.
5. (amended) [A] The method as claimed in claim 1, wherein [any one of the preceding claims, characterised in that] the induced cells containing the trehalose are dried prior to their use in the production of the vaccine composition.
6. (amended) [A] The method as claimed in claim 5, [characterised in that ] wherein the cells are dried in the absence of added extra-cellular carbohydrate glassy stabilising matrix.

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7. (amended) [A] The method as claimed in claim 1, wherein [any one of the preceding claims, characterised in that] the procaryotic cells are bacteria, protozoa or fungi.
8. (amended) [A] The method as claimed in claim 1, wherein [any one of the preceding claims, characterised in that] the procaryotic cells are treated by cultivating them in a medium containing one or more solutes and having an osmolarity of at least 350 mOsmoles.
9. (amended) [A] The method as claimed in claim 8, wherein [characterised in that] the solute is selected from [a] the group consisting of sodium, potassium, calcium and [/ or] ammonium [salt], and combinations thereof.
10. (amended) [A] The method as claimed in claim 1, wherein [characterised in that] the procaryotic cell has been modified so as to synthesise trehalose.
11. (amended) [A] The method as claimed in claim 1, wherein [characterised in that] the treatment of the cells is carried out to achieve a concentration of trehalose within the cells of at least 100mM.
12. (amended) [A] The method as claimed in claim 1, wherein [any one of the preceding claims, characterised in that] the procaryotic cells containing the induced trehalose are killed prior to use in the vaccine composition.
13. (amended) [A] The method as claimed in claim 1, wherein [any one of the preceding claims, characterised in that] the treatment of the procaryotic cells is carried out *in vitro*.
14. (amended) A vaccine composition comprising an immunogenic determinant, [characterised in that] wherein the immunogenic determinant includes a procaryotic cell or cell residue which contains at least 10mM of trehalose within the cell.

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15. (amended) A vaccine composition comprising [characterised in that it contains] an immunogenic determinant produced by the method of claim 1 [any of claims 1 to 13].
16. (amended) [A] The vaccine composition as claimed in claim 14, comprising [either of claims 14 or 15, characterised in that it contains] an adjuvant for the immunogenic determinant.
17. (amended) [A] The vaccine composition as claimed in claim 14, comprising [any one of claims 14 to 16, characterised in that it contains] an aqueous carrier.
18. (amended) A vaccine composition as claimed in claim 14, wherein [any one of claims 14 to 17, characterised in that] the induced cells containing trehalose [are] have been dried in the presence of a non-reducing carbohydrate to provide a storage stable but viable immunogenic determinant for storage prior to use in [a] the vaccine composition.
20. (amended) A method for treating an animal with a vaccine, comprising administering to said animal [characterised in that] a pharmaceutically effective amount of a vaccine composition as claimed in claim 1 [any one of claims 14 to 18 is administered to the animal] to elicit an immune response in the animal.
21. (amended) [A] The method as claimed in claim 20, wherein [characterised in that] the vaccine composition is administered by injection.
22. (amended) A prokaryotic cell which has had its genetic structure modified so as to remove or inhibit that portion of the genetic structure which inhibits or restricts the synthesis of trehalose by the cell, whereby the cell constitutively synthesises trehalose within the cell as it grows.